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Biochemical characterisation of the seed oils of four safflower (*Carthamus tinctorius*) varieties grown in north-eastern of Morocco

Abdessamad Ben Moumen,[1] Farid Mansouri,[1] Gaetan Richard,[2] Malika Abid,[1] Marie-Laure Fauconnier,[2] Marianne Sindic,[3] Ahmed El Amrani[1] & Hana Serghini Caid[1]*

1 Laboratoire Biologie des Plantes et des Microorganismes, Faculté des Sciences, Université Mohammed Premier, BP-717 Oujda, Morocco
2 Faculté Gembloux Agro-BioTech, Unité Chimie Général et Organique, Université de Liège, Passage des Deportes 2, B-5030 Gembloux, Belgium
3 Faculté Gembloux Agro-BioTech, Laboratoire qualité et sécurité Alimentaire, Unité qualité et risque, Université de Liège, Passage des Deportes 2, B-5030 Gembloux, Belgium

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Summary The quality of the oil of four safflower varieties, originating from Spain (Rancho), India (Sharda) and Morocco (Cartamar and Cartafri), which were cultivated at the experimental station in Oujda (a semi-arid region of eastern Morocco) was evaluated through analysis of their phenolic and carotenoid contents. The composition of the phenolic compounds of safflower oil has not yet been documented. Therefore, in this preliminary study, Thirty different phenolic compounds were identified, and significant differences between the oil varieties were observed ($P < 0.05$). In the seed oil from the Rancho and Sharda safflower varieties, the main phenolic compound was trans-chalcone, representing 13.45% and 11.8%, respectively, of the total phenolics, whereas in Cartamar and Cartafri oils, naringin accounted for 26.82% and 16.5%, respectively, of the total phenolics. The total carotenoid contents ranged from 1.13 mg kg^{-1} (Rancho) to 1.34 mg kg^{-1} (Cartamar and Cartafri). We observed that β-cryptoxanthin (0.31–0.37 mg kg^{-1}) and β-carotene (0.3–0.35 mg kg^{-1}) were the predominant carotenoids in all of the safflower oils that were studied.

Keywords Carotenoids, eastern Morocco, oil quality, phenolic compounds, safflower.

Introduction

The safflower (*Carthamus tinctorius* L) is an oilseed crop, which for many years has been grown in relatively small areas of northern and middle-eastern Africa (Purdy *et al*., 1959). The safflower is a tap-rooted annual crop that can tolerate environmental stresses, including salinity and water stresses (Lovelli *et al*., 2007). Therefore, this crop could have a good yield potential in Morocco's semi-arid areas. The Moroccan research of this crop mainly focused on its breeding, starting in the early 1960s (Nabloussi & Boujghagh, 2006).

Safflower oil (*Carthamus tinctorius* L.) has a high content of polyunsaturated linoleic acid and tocopherol and is produced for nutritional as well as for medicinal uses (Fernandez-Martinez *et al*., 1993). Secondary metabolites are a subject of increasing research interest. This is particularly true for plant polyphenols that are widely used in therapy due to their vasculoprotective, anti-inflammatory, enzyme-inhibitory, antioxidant and radical-scavenging properties (Bahorun *et al*., 1996).

The quality of vegetable oil depends on several factors, including the plant variety, the degree of its maturation, pedoclimatic factors, farming practices, the oil-extraction method utilised and the storage conditions (Matos *et al*., 2007). Safflower oil has been characterised in different regions of the world, and several experiments have demonstrated variability in its composition due to the soil and climatic conditions. Thus, the introduction of a new crop in a regional farming system such as that in north-eastern Morocco requires information concerning its performance under the local environmental conditions.

The aim of the present work was to complete our previous investigation focused on characterising

*Correspondent: Fax: 00 212 536 500 603;
e-mail: hanaserghini@hotmail.com

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safflower seeds produced in north-eastern Morocco (Ben Moumen *et al.*, 2013a,b). In addition to determining the fatty acid and triglyceride compositions, our main objective was to determine, for the first time, the precise phenolic and carotenoid contents of oils obtained from the seeds of four safflower varieties that were cultivated in north-eastern Morocco. The contents were compared with those of other safflower oils prepared from cultivants grown in other countries and with those of the principal oils produced in Morocco (olive and argan oils).

Materials and methods

Plant material

The plant material consisted of the following four *Carthamus tinctorius* varieties (pure lines) of different origins: 'Cartafri' and 'Cartamar' (Morocco), 'Rancho' (Spain) and 'Sharda' (India). Seeds of all these varieties were kindly provided by the National Institute for Agricultural Research (INRA), Regional Research Centre of Meknes (Morocco).

Cultivation was conducted at the Centre for Agricultural Qualification Bouchtat in Oujda (north-eastern Morocco).

Oil extraction

The seeds were ground using a coffee grinder. The Soxhlet method was used to extract oil 30 g of ground seed flour using 100% n-hexane for 5 h. The oils were recovered using a rotary evaporator and were stored in the dark at 6 °C. The results were expressed as the percentage of the lipids in the dry seed powder. All of oil extractions were performed in triplicate.

Analytical methods

Fatty acid analysis

The fatty acids were converted to fatty acid methyl esters before analysis by mixing a solution of 10 mg of oil in 0.2 mL of hexane with 0.5 mL of solution A (solution A: 55 mL of dry methanol + 20 mL of pentane + 25 mL of BF3 at 14% weight in methanol), placing the mixture in a water bath at 75 °C for 90 min and then adding 0.6 mL of saturated NaCl and 0.2 mL of 10% H_2SO_4 (V:V).

The fatty acid methyl esters were analysed using an HP 6890 series gas chromatography system equipped with a capillary column (Supelcowax: 30.0 m × 250 mm × 0.25 μm) and an FID detector. The carrier gas was nitrogen, supplied at a flow rate of 1.7 mL min^{-1}. The temperatures of the injector and detector were set at 150 and 250 °C, respectively, and the oven temperature was set at 210 °C. The injection volume was 1 μL.

Triglyceride analysis

The samples were analysed using high-performance liquid chromatography (HPLC). The Shimadzu LC-6AD HPLC system used consisted of a computer-controlled double plunger system connected to two LC-10AD Shimadzu liquid chromatography pumps and a refractive index detector (RID) 10A. A Kromasil C18 column (4.6 × 250 mm, particle size of 5 μm) was used. The isocratic solvent used as the mobile phase was composed of acetone/acetonitrile (60/40), delivered at a flow rate of 1 mL min^{-1}, and the sample injection volume was 20 μL. All of the separations were performed at ambient temperature. The elution order was a function of the number of carbon atoms and the degree of unsaturation.

Polyphenol extraction

The phenolic compounds were extracted according to the method described by Ollivier *et al.* (2004), with some modifications.

A 2.5-mL aliquot of a methanol/water solution (80/20. V/V) was added to 2.5 g of safflower oil in a centrifuge tube. After 10 min of vigorous mixing, the tubes were centrifuged for 15 min at 500 ***g***. The methanolic phase was recovered and transferred to a 5-mL volumetric flask. This operation was repeated two times and the volume was brought to 5 mL using the methanol/water solution (80/20 V/V).

Determination of the total phenolic content

A 2-mL aliquot of each solution was placed in a test tube, and 1 mL of Folin-Ciocalteu reagent (Sigma Aldrich, St Louis, MO, USA), 5 mL of distilled water and 5 mL of a 10% solution of sodium carbonate (Na_2CO_3) was added. The solutions were shaken immediately and were thoroughly mixed and then were maintained in darkness for 30 min. The absorbance of each solution at 760 nm relative to that of a blank was determined. A calibration curve was obtained using four solutions of caffeic acid at concentrations of 0.02–0.28 mg mL^{-1}.

Analysis of phenolic compounds by HPLC

The phenolic compounds were separated using a Zorbax SB-C18 column (4.6 × 250 mm, particle size of 5 μm). An Agilent 1100 pump equipped with an automatic injector was used. The solvent system used was a gradient of A (water with 0.5% formic acid) and B (methanol), supplied at a flow rate of 1 mL min^{-1}. The column was equilibrated for at least 15 min using the elution solvent before the analysis was begun. Detection was performed using a UV-visible detector set at 280 nm. A 10-μL aliquot of the final sample

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solution was injected into the HPLC system. The compounds were identified by comparing their elution times with those of standards. For this analysis, standard solutions of the following 20 polyphenolic compounds were prepared in methanol: gallocatechin, epigallocatechin, epigallocatechin gallate, naringin, rutin hydrate, hydroxytyrosol, tyrosol, 4-hydroxyphenyl lactic acid, vanillic acid, syringic acid, ferulic acid, p-coumaric, cinnamic acid, benzoic acid, sinapic acid, vanillin, 1.3-dihydroxynaphthalene, luteolin, pinoresinol, apigenin and quercetin (all obtained from Sigma Aldrich).

Determination of the content of different carotenoids

The procedure described by Zakaria *et al*. (1979) was used to extract the carotenoids from the oil samples. The content of different carotenoids in the extracts was determined through measuring the absorbance using a spectrophotometer and applying the following extinction coefficients for $E_{1cm}^{1\%}$ in petroleum ether (60–80 °C): for α-carotene, 2710 at 445 nm; for β-carotene, 2500 at 450 nm; for γ- carotene, 2720 at 461 nm and for β-cryptoxanthin, 2386 at 452 nm.

Statistical analysis

All of the analytical determinations were performed at least in triplicate. The values of the different parameters were expressed as the mean value ± standard deviation (SD). Significant differences between the mean values (at $P < 0.05$) were determined using an ANOVA using SPSS software for Windows (SPSS ver. 19, Chicago, IL, USA).

Results and discussion

Oil contents and oil-quality parameters

The content of an oil is not a criterion that determines its quality, but is rather a norm to be considered during varietal selection (Allalout *et al*., 2009). In a varietal trial study, the safflower oil contents (by Soxhlet method) were found to range from 27 to 40%, indicating a significant potential for cultivar improvement through plant selection (Mailer *et al*., 2008). Among the seeds of the four varieties of safflower, the oil content of Rancho seeds (Table 1) was the highest, at 35.38 g/100 g of seeds, followed by Cartafri seeds, which contained 31.63 g of oil/100 g of seeds, Sharda seeds, which contained 30.43 g of oil/100 g of seeds and Cartamar seeds, which contained 28.81 g of oil/100 g of seeds. These values are similar to those reported previously (Ashrafi & Razmjoo, 2010; Gecgel *et al*., 2007). The characterisation data for these oils are shown in Table 1 (Ben Moumen *et al*., 2013a,b). The free fatty acid (FFA) content or acid value is one of the most important parameters in defining the quality of oil, and it is often determined to classify and/or evaluate oil. The FFA value is an indicator of the extent of the hydrolysis of triglycerides to produce diglycerides and monoglycerides, which liberates free fatty acids. All of the analysed oils had very low contents of free fatty acids, with the *Rancho* and *Sharda* oils containing 0.7% FFA, the *Cartamar* oil containing 0.63% FFA and the Cartafri oil containing 0.67% FFA.

The peroxide value of the oil samples ranged from 4.58 to 31.21 meq O2 per Kg (Table 1). Three of the oil samples studied exhibited a peroxide value that was lower than the maximum limit of 20 meq O2 per Kg established for extra virgin olive oils. A higher peroxide value (31.21 meq O_2 Kg^{-1}) was found in the oil derived from the seeds of the Spanish variety *Rancho*.

The PV results were relatively high compared with those reported in the literature (Rafiquzzaman *et al*., 2006). These results do not necessarily indicate the deterioration of the oils within the seeds, but might be explained by the method of oil-extraction utilised. Indeed, the Soxhlet method used in this study has the drawback of being a hot extraction procedure that promotes lipid peroxidation. Using a cold-extraction method, (Bligh & Dyer, 1959) led to *Cartafri* oil with a peroxide value of 6.6 meq O_2 Kg^{-1} (data not shown) instead of 15.2 meq O_2 Kg^{-1}.

Fatty acid composition

Table 2 shows the relative proportions of the various fatty acid methyl esters (FAMEs) in the oils. Palmitic, oleic, stearic and linoleic esters were the main FAMEs identified. Saturated fatty acids represented 10.8% to 9.7% of FAs, with the majority being palmitic acid (7.2–8.6%) and stearic acid (2.0–2.4%). The content of unsaturated fatty acids in the oils was high, at 77.94% and 79.98% for the Cartamar and Sharda oils, respectively. The unsaturated fatty acids were principally represented by linoleic acid (77.9–79.5%) and oleic acid (9.5–11.3%). Environmental, mainly temperature, effects on the fatty acid profiles of safflower oils have been demonstrated; however, this study showed that the fatty acid composition of the safflower oils obtained from the seeds of plants cultivated in north-eastern Morocco was similar to those observed by other authors in several countries (Rafiquzzaman *et al*., 2006; Bozan & Temelli, 2008) but differed from those of oils obtained from plants cultivated in Turkey (Aydeniz *et al*., 2014) or in Iran (Yeilaghi *et al*., 2012).

Triacylglycerol composition

After determining the fatty acid composition of the safflower oils, we studied the possible combinations of acyl chains on the glycerol backbone.

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Table 1 Physicochemical quality of safflower oils samples

	Varieties			
Physicochemical parameters	**Rancho**	**Cartamar**	**Cartafri**	**Sharda**
Free Acidity (%)	0.70 ± 0.03^{b}	0.63 ± 0.05^{a}	0.67 ± 0.04^{ab}	0.70 ± 0.02^{b}
Peroxide value (meq O2 Kg^{-1})	31.21 ± 1.41^{c}	4.58 ± 1.04^{a}	15.64 ± 1.59^{ab}	15.81 ± 3.73^{ab}
Oil content (g/100 g)	33.84 ± 1.51^{c}	27.67 ± 1.06^{a}	30.55 ± 0.94^{b}	29.35 ± 0.93^{ab}

Values are the means of the four different safflower oils samples (n = 3) ± standard deviations. Significant differences in the same row are shown by different letters (a–c) varieties (P < 0.05).

Table 2 Fatty acid compositions (% GC area) for Rancho, Cartamar, Cartafri and Sharda monovarietal safflower oils

	Varieties			
Fatty acid %	**Rancho**	**Cartamar**	**Cartafri**	**Sharda**
14: 0	0.12 ± 0.01^{a}	0.13 ± 0.02^{ab}	0.16 ± 0.02^{b}	0.15 ± 0.03^{ab}
16: 0	7.36 ± 0.38^{a}	7.20 ± 0.52^{a}	8.60 ± 0.45^{b}	8.12 ± 0.9^{ab}
16: 1	0.08 ± 0.00^{b}	0.09 ± 0.01^{b}	ND	0.10 ± 0.01^{c}
18: 0	2.19 ± 0.00^{b}	2.39 ± 0.09^{c}	2.00 ± 0.14^{a}	2.15 ± 0.06^{ab}
18: 1	10.63 ± 0.09^{b}	11.29 ± 0.14^{c}	10.61 ± 0.31^{a}	9.50 ± 0.37^{ab}
18: 2	79.49 ± 0.48^{b}	77.94 ± 0.33^{c}	78.62 ± 0.02^{a}	79.98 ± 1.24^{ab}
18: 3	ND	0.09 ± 0.00^{b}	ND	ND
20: 0	ND	0.19 ± 0.05^{b}	ND	ND
22: 0	ND	0.16 ± 0.04^{b}	ND	ND
∑SFAs	9.67	10.14	10.76	10.42
∑MUFAs	10.71	11.82	10.61	9.60
∑PUFA	79.49	77.94	78.62	79.98

Values are the means of four different safflower oils samples ± standard deviations. (ND, not determined; SFA, saturated fatty acid; MUFA, monounsaturated fatty acid; PUFA, polyunsaturated fatty acid. ∑, sum; 14:0, myristic acid; 16:0, palmitic acid; 16:1, palmitoleic acid; 18:0, stearic acid; 18:1, oleic acid; 18:2, linoleic acid; 18:3, linolenic acid; 20:0, arachidic acid and 22:0, behenic acid). Significant differences in the same row are shown by different letters (a – c) varieties (P < 0.05).

Analysis of the molecular species of triacylglycerol (TAG) in the oils using HPLC-RID (Table 3) distinguished ten main molecular TAG species, as follows: LLL, LPL, LLO, PLO, LLS, POP, OOO, POO, SOO and PPL (with L corresponding to a linoleate moiety, P to a palmitate moiety, O to an oleate moiety and S to a stearate moiety). Three major species of TAGs represented more than 80% of the total TAGs in the oils of all four varieties. The major peak occurred at ECN (equivalent carbon number) 42, which corresponded to trilinolein (LLL). The amount of this triglyceride in the oils ranged from 52.4% (*Cartafri*) to 56.5% (*Rancho*). We have conducted a thorough literature search and no information, in authentic scientific references, on molecular species of triglycerides could be found.

Phenolic analyses

Vegetable oils are composed mainly of triacylglycerol (95–98%) and complex mixtures of minor compounds (2–5%) of a wide range of chemical natures. The qualitative and quantitative features of these minor constituents differ greatly, depending on the vegetal species from which the oils were purified. Moreover, for the same species, the content and composition of these components may vary due to agronomic and climatic conditions, the quality of the fruits or seeds and the oil-extraction and refining procedures used.

Total phenolic content

Significant differences in the total phenolic contents (determined by the Folin-Ciocalteu method) of the studied oil varieties were found (P < 0.05). The total phenolic content of the four studied varieties of oils (Table 4) was highest in *Rancho* oil (143.7 mg kg^{-1}), followed by Cartamar oil, with 125. 8 mg kg^{-1} of phenolics and then by Cartafri and Sharda oils, which contained 118.2 and 97.5 mg kg^{-1} of phenolics, respectively. These observed values were similar to those reported by Mailer *et al.* (2008).

Considering the high consumer demand for oils rich in phenolics, we compared the phenolic contents of the safflower oils to those of the principal oils produced in Morocco. The total phenolic content of all of the

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Table 3 Triacylglycerol molecular species of Rancho, Cartamar, Cartafri and Sharda monovarietal safflower oils

	Varieties			
Triacylglycerol %	**Rancho**	**Cartamar**	**Cartafri**	**Sharda**
LLL	56.53 ± 0.12[d]	54.97 ± 0.07[c]	52.43 ± 0.14[a]	54.05 ± 0.04[b]
LLO	19.49 ± 0.31[c]	17.97 ± 0.04[a]	20.96 ± 0.15[d]	19.14 ± 0.01[b]
LLP	13.18 ± 0.03[b]	12.54 ± 0.18[a]	13.72 ± 0.35[c]	12.47 ± 0.03[a]
PLO	0.11 ± 0.04[a]	0.44 ± 0.02[b]	0.37 ± 0.18[b]	0.39 ± 0.01[b]
LLS	0.10 ± 0.05[a]	3.21 ± 0.02[b]	3.70 ± 0.08[d]	3.43 ± 0.08[c]
PPL	2.47 ± 0.03[a]	4.51 ± 0.02[b]	4.36 ± 0.27[b]	4.38 ± 0.01[b]
OOO	4.08 ± 0.10[c]	3.16 ± 0.10[a]	3.47 ± 0.04[b]	3.40 ± 0.02[b]
POO	2.23 ± 0.09[a]	0.45 ± 0.07[ab]	0.51 ± 0.03[b]	0.38 ± 0.01[a]
POP	0.50 ± 0.02[b]	0.47 ± 0.06[b]	0.05 ± 0.03[a]	0.48 ± 0.01[b]
SOO	1.08 ± 0.11[b]	1.07 ± 0.03[b]	0.13 ± 0.03[a]	1.45 ± 0.09[c]

L, Linoleate; O, Oleate; S, Stearate; P, Palmitate.
Values are the means of the four different safflower oils samples ($n = 3$) ± standard deviations. Significant differences in the same row are shown by different letters (a – c), varieties ($P < 0.05$).

Table 4 Total Phenols content (mg kg^{-1}) and phenols composition (%) of Rancho, Cartamar, Cartafri and Sharda monovarietal safflower oils

		Varieties			
Pic	**Phenolic compounds %**	**Rancho**	**Cartamar**	**Cartafri**	**Sharda**
1	Tyrosol	ND	0.04 ± 0.00[b]	0.32 ± 0.02[c]	ND
2	Benzoic Acid	ND	0.26 ± 0.00[b]	0.43 ± 0.01c	ND
3	Vanillic Acid	0.33 ± 0.02[a]	0.48 ± 0.02[b]	0.57 ± 0.10[b]	0.81 ± 0.04[c]
5	Vanillin	3.34 ± 0.11[b]	1.88 ± 0.27[a]	4.78 ± 0.11[c]	5.53 ± 0.11[d]
7	(p) coumaric Acid	1.05 ± 0.02[c]	ND	ND	0.48 ± 0.04[b]
8	Sinapic Acid	ND	1.52 ± 0.09[c]	ND	0.78 ± 0.07[b]
9	Trans-ferulic Acid	0.85 ± 0.15[b]	ND	1.92 ± 0.14[c]	ND
10	Naringin	10.55 ± 1.18[b]	26.82 ± 1.32[d]	16.5 ± 0.74[c]	ND
11	Rutin	ND	ND	13.86 ± 0.13[b]	ND
13	1-3 DHN	0.49 ± 0.07[b]	2.00 ± 0.06[d]	0.88 ± 0.02[c]	0.29 ± 0.03[a]
14	Pinoresinol	0.55 ± 0.04[a]	3.34 ± 0.13[d]	1.14 ± 0.06[c]	0.83 ± 0.09[b]
17	Trans-cinnamic Acid	0.92 ± 0.18[c]	ND	ND	0.38 ± 0.02[b]
18	Trans-chalcon	13.45 ± 0.29[d]	8.03 ± 0.37[b]	ND	11.8 ± 0.12[c]
19	Phenol content (mg kg^{-1})	143.70 ± 27.92[b]	125.80 ± 5.28[ab]	118.20 ± 15.76[ab]	79.50 ± 15.87[a]

Values are the means of the four different safflower oils samples ($n = 3$) ± standard deviations. Significant differences in the same row are shown by different letters (a–d) varieties ($P < 0.05$). ND, not determined.

analysed samples was higher than that of argan oil, which does not exceed 89 mg kg^{-1} (Demnati *et al.*, 2011) but lower than that of virgin olive oil (209 mg kg^{-1}) (Mansouri *et al.*, 2013).

Phenolic compound contents

Analysis of the phenolic compounds in the four varieties of safflower oils using reversed phase HPLC (Table 4) led to the identification of 13 phenolic compounds. When possible, the retention times of the sample peaks were compared to those of available reference compounds that were analysed under the same LC conditions. Thus, it was possible to identify two hydroxybenzoic acids (vanillic acid and syringic acid), four hydroxycinnamic acids (*p*-coumaric acid, cinnamic acid, sinapic acid and ferulic acid) and five compounds from other phenolic classes (1-3 DHN, pinoresinol, vanillin, rutin, trans-chalcone, naringin and tyrosol) in the four oil varieties studied.

Significant differences among the phenolic compositions of the oil varieties were observed ($P < 0.05$). The main phenolic compound in Rancho and Sharda oils was *trans*-chalcone (13.45% and 11.8%, respectively), whereas that of Cartamar and Cartafri oils was naringin (respectively, 26.82% and 16.5%).

Only a few studies that were specifically devoted to the separation and identification of phenolic compounds in safflower oil are available. Yu *et al.* (2013)

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Table 5 Carotenoids composition (mg kg^{-1}) for Rancho, Cartamar, Cartafri and Sharda monovarietal safflower oils

	Varieties			
Carotenoids mg kg^{-1}	Rancho	Cartamar	Cartafri	Sharda
α carotene	0.28 ± 0.03^{a}	0.32 ± 0.08^{a}	0.32 ± 0.00^{a}	0.28 ± 0.13^{a}
β carotene	0.30 ± 0.08^{a}	0.35 ± 0.09^{a}	0.35 ± 0.01^{a}	0.31 ± 0.14^{a}
γ carotene	0.25 ± 0.11^{a}	0.30 ± 0.09^{a}	0.29 ± 0.01^{a}	0.27 ± 0.11^{a}
β cryptoxanthin	0.31 ± 0.08^{a}	0.37 ± 0.09^{a}	0.36 ± 0.01^{a}	0.32 ± 0.15^{a}
Σ analysed carotenoids	1.14 ± 0.34^{a}	1.34 ± 0.34^{a}	1.32 ± 0.02^{a}	1.18 ± 0.54^{a}

Values are the means of the four different safflower oils samples (n = 3) ± standard deviations. Significant differences in the same row are shown by different letters (a–d) varieties (P < 0.05).

showed that the major phenolic compounds in a hot-water extract of *Carthamus tinctorius* L seeds were (−) epigallocatechin (109.62 mg g^{-1}), a 4-hydrox ybenzohydrazide derivative (18.28 mg g^{-1}) and gallocatechin (17.02 mg g^{-1}).

Carotenoid content

The main pigments present in edible oils are carotenoids (Serani & Piacenti, 1992). In general, the carotenoid content of oil is an important quality parameter due to the correlation between the colour of the oil, its carotenoid content, and the evaluation of its quality (Aparicio *et al*., 1999). Carotenes, one of the two classes of carotenoids, are essential antioxidant nutrients that protect oils against oxidative damage (Luterotti *et al*., 2002).

Four different carotenoids were analysed, as follows: α carotene, β carotene, γ carotene and β cryptoxanthin (Table 5). Considering the total carotene contents, no significant differences between the oil varieties was observed (P < 0.05). The content of total carotenoids ranged from 1.13 mg kg^{-1} (Rancho) to 1.34 mg kg^{-1} (Cartamar and Cartafri). We observed that β cryptoxanthin (0.31–0.37 mg kg^{-1}) and β-carotene (0.3–0.35 mg kg^{-1}) were the predominant carotenoids in all of the safflower oils studied.

Compared to virgin olive oil, which contains between 1.0 and 4.2 mg kg^{-1} of carotenoids (Baccouri *et al*., 2008) and argan oil, in which the carotenoid concentration can reach 10–25 mg kg^{-1} (Matthäus *et al*., 2010), the studied safflower oils had low levels of total carotenoids. However, the wealth of β cryptoxanthin that distinguishes safflower oil is an important asset because β cryptoxanthin prevented adipocyte hypertrophy by downregulating the expression of peroxisome proliferator-activated receptor gamma (PPAR-γ) via RAR, suppressed mastocytosis (Takayanagi & Mukai, 2014). Study with β cryptoxanthin (0.75 mg per day) and phytosterols (1.5 g per day) combined was performed in 38 postmenopausal women improved the cholesterol-lowering effect of phytosterols; additionally this combination may also be beneficial in reducing the risk of osteoporosis (Granado-Lorencio *et al*., 2014).

Conclusions

The phenolic compound composition of safflower oil has not been previously reported documented. Therefore, in this preliminary study, 13 different phenolic compounds were identified for the first time, and significant differences in terms of the quality and quantity of the oil varieties were observed (P < 0.05). Our results showed that safflower oils obtained from the seeds of plants cultivated in north-eastern Morocco were rich in phenolic constituents and demonstrated good antioxidant activity (Ben Moumen *et al*., 2013a, b). Thus, this oil, being rich in phenolic acids, could be a good source of natural antioxidants, and safflowers could be an alternative oilseed crop in eastern Morocco (in the marginal areas), where the only source of oil is olives, which are expensive and cannot satisfy the need for oil.

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References

Allalout, A., Krichène, D., Methenni, K. *et al*. (2009). Characterization of virgin olive oil from super intensive Spanish and Greek varieties grown in northern Tunisia. *Scientia Horticulturae*, **120**, 77–83.

Aparicio, R., Roda, L., Albi, M.A. & Gutiérrez, F. (1999). Effect of various compounds on virgin olive oil stability measured by Rancimat. *Journal of Agricultural and Food Chemistry*, **47**, 4150–4155.

Ashrafi, E. & Razmjoo, K. (2010). Effect of irrigation regimes on oil content and composition of safflower (*Carthamus tinctorius* L.) cultivars. *Journal of the American Oil Chemists' Society*, **87**, 499–506.

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Aydeniz, B., Güneşer, O. & Yılmaz, E. (2014). Physico-chemical, sensory and aromatic properties of cold press produced safflower oil. *Journal of the American Oil Chemists' Society*, **91**, 99–110.

Baccouri, B., Zarrouk, W., Baccouri, O. *et al*. (2008). Composition, quality and oxidative stability of virgin olive oils from some selected wild olives. J. *Grasas y aceites*, **59**, 346–351.

Bahorun, T., Gressier, B., Trotin, F. *et al*. (1996). Oxygen species scavenging activity of phenolic extracts from hawthorn fresh plant organs and pharmaceutical preparations. *Arzneimittel-Forschung*, **46**, 1086–1089.

Ben Moumen, A., Zraibi, L., Mansouri, F. *et al*. (2013a). Chemical composition and antioxidant activity of seed oils of four safflower (*Carthamus tinctorius*) varieties grown in eastern Morocco. In: Symposium International des Plantes Aromatiques et Médicinales (SIPAM5). Marrakech.

Ben Moumen, A., Mansouri, F., Zraibi, L. *et al*. (2013b). Comparative study of four safflower oils (*Carthamus tinctorius*) varieties grown in eastern of Morocco. In Book of Proceedings InsideFood Symposium).

Bligh, E.G. & Dyer, W.J. (1959). A rapid method of total lipid extraction and purification. *Canadian Journal of Biochemistry and Physiology*, **37**, 911–917.

Bozan, B. & Temelli, F. (2008). Chemical composition and oxidative stability of flax, safflower and poppy seed and seed oils. *Bioresource Technology*, **99**, 6354–6359.

Demnati, D., Sánchez, S., Pacheco, R., Zahar, M. & Martínez, L. (2011). Comparative study of argan and olive fruits and oils. *Actes du premier congrés international de l'arganier, Agadir 15 - 17 décembre 2011*.

Fernandez-Martinez, J., Del Rio, M. & De Haro, A. (1993). Survey of safflower (*Carthamus tinctorius* L.) germplasm for variants in fatty acid composition and other seed characters. *Euphytica*, **69**, 115–122.

Gecgel, V., Demirci, M. & Esendal, E. (2007). Seed yield, oil content and fatty acids composition of safflower (*Carthamus tinctoruis* L.) varieties sown in spring and winter. *International Journal of Engineering Science*, **1**, 11–15.

Granado-Lorencio, F., Lagarda, M.J., Garcia-Lopez, F.J. *et al*. (2014). Effect of beta-cryptoxanthin plus phytosterols on cardiovascular risk and bone turnover markers in post-menopausal women: a randomized crossover trial. *Nutrition, Metabolism, and Cardiovascular Diseases*, **24**, 1090–1096.

Lovelli, S., Perniola, M., Ferrara, A. & Di Tommaso, T. (2007). Yield response factor to water (Ky) and water use efficiency of *Carthamus tinctorius* L. and *Solanum melongena* L. *Agricultural Water Management*, **92**, 73–80.

Luterotti, S., Franko, M., Šikovec, M. & Bicanic, D. (2002). Ultrasensitive assays of *trans*-and *cis*-β-carotenes in vegetable oils by high-performance liquid chromatography–thermal lens detection. *Analytica Chimica Acta*, **460**, 193–200.

Mailer, R., Potter, T., Redden, R. & Ayton, J. (2008). Quality evaluation of safflower (*Carthamus tinctorius* L.) cultivars. In: 7th International Safflower Conference, Wagga Wagga, NSW. Pp. 3–6.

Mansouri, F., Ben Moumen, A., Lopez, G. *et al*. (2013). Preliminary Characterization of monovarietal virgin olive oils produced in eastern area of Morocco. In: *Book of Proceedings InsideFood Symposium*.

Matos, M., Pérez-Flores, V., Camacho, M., Pernaute, B., Pinto-Carnide, O. & Benito, C. (2007). Detection and mapping of SSRs in rye ESTs from aluminium-stressed roots. *Molecular Breeding*, **20**, 103–115.

Matthäus, B., Guillaume, D., Gharby, S., Haddad, A., Harhar, H. & Charrouf, Z. (2010). Effect of processing on the quality of edible argan oil. *Food Chemistry*, **120**, 426–432.

Nabloussi, A. & Boujghagh, M. (2006). Safflower breeding in Morocco: history and current situation. *Sesame and Safflower Newsletter*, **21**, 84–87.

Ollivier, D., Boubault, E., Pinatel, C., Souillal, S., Guérére, M. & Artaud, J. (2004). Analyse de la fraction phenolique des huiles d'olive vierges. J. *Annales des falcifications*. *De l'expertise chimique et toxicologique*, **965**, 169–196.

Purdy, R., Cummings, L., Claassen, C. & Kneeland, J. (1959). Safflower, its development and utilization. *Journal of the American Oil Chemists Society*, **36**, a26–a30.

Rafiquzzaman, M., Hossain, M.A. & Hasan, A.M. (2006). Studies on the characterization and glyceride composition of safflower (*Carthamus tinctorius*) seed oil. *Bangladesh Journal of Scientific and Industrial Research*, **41**, 235–238.

Serani, A. & Piacenti, D. (1992). Kinetics of pheophytin-A photodecomposition in extra virgin olive oil. *Journal of the American Oil Chemists' Society*, **69**, 469–470.

Takayanagi, K. & Mukai, K. (2014). Chapter 34 – Beta-Cryptoxanthin, a Novel Carotenoid Derived from Satsuma Mandarin, Prevents Abdominal Obesity. In: *Nutrition in the Prevention and Treatment of Abdominal Obesity* (edited by R.R. Watson). Pp. 381–399. Burlington: Elsevier Science.

Yeilaghi, H., Arzani, A., Ghaderian, M., Fotovat, R., Feizi, M. & Pourdad, S.S. (2012). Effect of salinity on seed oil content and fatty acid composition of safflower (*Carthamus tinctoriu*s L.) genotypes. *Food Chemistry*, **130**, 618–625.

Yu, S.-Y., Lee, Y.-J., Kim, J.-D. *et al*. (2013). Phenolic composition, antioxidant activity and anti-adipogenic effect of hot water extract from safflower (*Carthamus tinctorius* L.) seed. *Nutrients*, **5**, 4894–4907.

Zakaria, M., Simpson, K., Brown, P.R. & Krstulovic, A. (1979). Use of reversed-phase high-performance liquid chromatographic analysis for the determination of provitamin A carotenes in tomatoes. *Journal of Chromatography A*, **176**, 109–117.

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